IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No.	:	**10/782,075**
Applicants	:	**Sean D. Monahan et al.**
Filed	:	**02/19/2004**
Art Unit	:	**1635**
Examiner	:	**Chong, Kimberly**
Docket No.	:	**25772 US2**

For: **Covalent Modification of RNA for In Vitro and In Vivo Delivery**

Commissioner of Patents
PO Box 1450
Alexandria, VA 22313-1450

APPELLANT'S BRIEF

TABLE OF CONTENTS

		Page(s)
	Table of Contents	2
i.	Real party in interest	3
ii.	Related appeals and interferences	4
iii	Status of Claims	5
iv.	Status of amendments	6
v.	Summary of claimed subject matter	7
vi.	Grounds of rejection to be reviewed on appeal	8
vii.	Argument	9
viii.	Claims appendix	14
ix.	Evidence appendix	16
x.	Related proceedings appendix	17
	Signature page	18

i. Real party in interest:

The real parties in interest are: Sean D. Monahan, Vladimir G. Budker, Lisa Nader, Vladimir Subbotin, and Jon A. Wolff, and, by assignment, Mirus Bio Corporation, which has changed its name to Roche Madison Inc. and then to Arrowhead Madison Inc, incorporated under the laws of the State of Delaware and located at 465 Science Drive, Suite C, Madison, WI 53711. Arrowhead Madison Inc. is a wholly owned subsidiary of Arrowhead Research Corporation, a California corporation, having its principal place of business in Pasadena, California, USA.

ii. Related appeals and interferences:

There are no interferences known to appellant, the appellant's legal representative, or assignee which will directly affect or be directly affected by or have a bearing on the Board's decision in the pending appeal.

iii. Status of Claims:

Claims 1, 4-6, 10, 13, and 14 have been rejected and are hereby appealed.

Claims 4-6, 10, 13, and 14 stand or fall with claim 1.

iv. Status of amendments:

No Amendments have been filed subsequent to the appealed rejection.

v. Summary of claimed subject matter for independent claim 1:

Applicants have invented a method and compositions for delivering RNAs to mammalian cells. Transfection reagents, polymer- and lipid-based, are used in the art to delivery nucleic acids into mammalian cells. Prior to Applicants' invention, interaction of the transfection reagent with the nucleic acid has relied on electrostatic interaction between the inherent negative charge of the nucleic acid and positive charge incorporated into the transfection reagent, either through the use of cationic polymers or cationic lipids. This electrostatic interaction results in reasonably stable complexes when used with large DNA's because of the length and thus high number of negative charges associated with the DNA. However, for certain small nucleic acids, such as siRNA and micro RNA (claim 10), which are as small as 20 base pairs or fewer, association with a transfection reagent through electrostatic interaction is very weak. Applicants have found, that by modifying the RNA to reversibly attach a hydrophobic group and selecting an amphipathic transfection reagent (one having both hydrophilic and hydrophobic parts), the RNA can be associated with the transfection reagent via hydrophobic interactions (Page 3 lines 24-32, Page 4 lines 1-2, Page 4 lines 8-11, Page 9 line 10 to page 10 line 14) in addition to or in lieu of electrostatic interactions. Because the hydrophobic groups is attached via labile bond cleavable under mammalian physiological conditions, the modification does not interfere with the function of the RNA once the RNA has been delivered to the cell (Page 2 lines 15-20, Page 5 lines 21-22). The transfection complexes thus formed are more efficient at delivery of RNA to the mammalian cell. The modified RNAs are also potentially more resistant to nuclease digestion (claim 13).

vi. Grounds of rejection to be reviewed on appeal:

Whether claims 1, 5, 6, 10, 13, and 14 are unpatentable under 35 U.S.C. 102(e) as being unpatentable over Fosnaugh et al. (U.S. 2003/0143732) as evidenced by Thierry et al. (US 6,110,490).

Whether claims 1, 4-6, 10, 13, and 14 are unpatentable under 35 U.S.C. 103(a) as being unpatentable over Fosnaugh et al. (U.S. 2003/0143732) taken with Manoharan, M. (Biochimica et Biophysica Acta 1489, 1999: 117-130) and further evidenced by Thierry et al (US 6,110,490).

vii. Argument:

a) Rejection under 35 U.S.C. 102(e) over Fosnaugh et al. (U.S. 2003/0143732, '732) as evidenced by Thierry et al. (US 6,110,490).

Applicants agree that '732 teaches a) an RNA can be modified b) the modification can be labile and that c) RNA can be delivered using a liposome vehicle. The Examiner concludes, therefore, the composition taught by '732 inherently possesses all the properties and functions of Applicants' claimed invention and thus '732 teaches labile linkage of a hydrophobic group to an RNA and specific use of this hydrophobic properties of the modified RNA to enhance interaction with a transfection reagent.

It is the Applicants' opinion that while all the words (labile modification of RNA and use of a transfection reagent which possesses hydrophobic character) are present in the Fosnaugh et al. disclosure, Applicants' invention is neither understood nor taught by '732 so as to be intelligently reproduced. '732 does not teach a composition formed by the association of an RNA to which a hydrophobic group is reversibly attached with an amphipathic transfection reagent wherein the modified RNA interacts with the amphipathic transfection reagent via hydrophobic interaction between the hydrophobic group linked to the RNA and a hydrophobic part of the amphipathic transfection reagent.

'732 is first a foremost an invention of a siRNA capable of inhibiting the adenosine A1 receptor (abstract, [0002]). To this end, '732 provides a staggering array of possible modifications and combinations, essentially providing a laundry list of possible modifications and methods of delivery known in the art, for this adenosine A1 receptor siRNA. '732 provides no coherent teaching with respect to which modifications are appropriate under which circumstances or which modifications are suitable with which delivery vehicles. A PHOSITA, on reading '732, would have no understanding as to which modifications, alone or in some further unknown combination, would have any particular desired characteristic. Except for an siRNA for inhibiting the adenosine A1 receptor (ADORA1) gene expression, the art of siRNA delivery is not advanced by the teaching of '732.

'732 teaches that siRNA can be modified and that the modification may be to a ribose, a nucleotide base, or to a phosphate backbone linkage [0026, 0167]. '732 specifically lists more than 150 modifications [0018, 0024, 0034, 0037, 0040, 0050, 0061, 0068, 0067, 0068, 0109, 0168, 0171, 0172, 0181, 0182, 0187, 0191] and repeatedly states that the specifically recited modifications are non-limiting examples and that *any other* nucleotide base modification is encompassed. While '732 further states that each of these modifications should not affect interaction of siRNA with a target RNA and/or other factors [0103], the authors provide no teaching as to which modifications will or will not affect this interaction.

'732 teaches that the modifications, including conjugates, may be used to: increase resistance to nuclease degradation, improve cellular uptake, improve stability of the interaction with the target, improve stability of the interaction with itself, enhance affinity and specificity to nucleic acid targets, overcome potential limitations of in vivo stability and bioavailability, provide longer half-life in serum, target particular cells or tissues, module polymerase activity, improve RNAi activity against ADORA1, enhance helical thermal stability, enhance shelf life, enhance half-life in vitro, enhance stability, or ease introduction to target site [0026, 0034, 0035, 0068, 0097, 0099, 0101, 0104, 0109, 0170, 0171, 0192]. Each of these is merely a desired effect. '732 does not teach which modifications may be used to affect which activities, properties, or characteristics. Particularly, '732 does not teach any modification which may be used to provide any interaction with any transfection reagent.

'732 teaches that conjugates and complexes can be used for delivery of biologically active molecules, (small molecules, lipids, phospholipids, nucleosides, nucleotides, nucleic acids, antibodies, toxins, negatively charged polymers, peptides, hormones, carbohydrates, polyethylene glycols, polyamines, proteins, and other polymers) across membranes [0172]. '732 does not teach or suggest which conjugate molecules or complexes can be used for delivery of which biologically active molecules across membranes or which of the conjugates and complexes is suitable with any of the listed biologically active molecules.

'732 teaches that transporters (e.g. conjugates) used to transport molecules across membranes may be used individually or as part of a multi-component system, may be used with or without degradable linkers, and may or may not be linked to the biologically active molecule

by a biodegradable linker [0172]. '732 does not teach or suggest *which* transporters should be used individually or as part of a multi-component system, *which* transporters may be used with degradable linkers, *which* transporters should be used without degradable linkers, or *which* transporter should be linked to the biologically active molecule and *which* cannot. Nor does '732 teach under which circumstance or conditions each of these unspecified transporters should or should not be part of a multi-component system, contain a biological linker or be linked to a biologically active molecule. Further, '732 does not teach any specific use for a biodegradable linkage, merely that such linkages may be used in the nucleic acid backbone, nucleic acid sugar, or nucleic acid base modifications [0173].

'732 teaches that conjugates and/or complexes of siRNA molecules may be used to facilitate delivery, transfer therapeutic compounds across cellular membranes, alter pharmacokinetics, or alter module localization [0172]. '732 does not teach which conjugates or complexes are suitable for each of these desired outcomes.

'732 teaches that the siRNA can be added directly to, complexed with, or mixed with: cationic lipids, liposomes, surface-modified liposomes, pH sensitive liposomes, pharmaceutically acceptable formulations, immunoliposomes, carriers, diluents, hydrogels, cyclodextrins, nanoparticles, biodegradable nanocapsules, microshperes, bioadhesive microspheres, proteinaceous vectors, fusogenic peptides, stabilizer, buffer, biopolymers, biodegradable polymers, troches, lozenges, aqueous suspensions, oily suspensions, dispersible powders or granules, emulsion, syrups, elixirs, *other* delivery vehicles, or otherwise delivered to target cells or tissues [0124, 0193, 0194, 0195, 0199, 0200, 0203]. '732 teaches that these vehicles may be used for storage or administration [0201]. '732 provides no teaching or direction for choosing a particular vehicle or choosing any modification in combination with any particular vehicle.

'732 teaches the use of liposomes for delivery of the siRNA. At [0195] '732 states, "When it is desired to use a liposome delivery mechanism, *standard protocols* for formation of liposomes can be followed." While liposomes are amphipathic transfection reagents as instantly claimed, it is *not* standard in the art to modify a nucleic acid to add a hydrophobic group prior to formation of the liposome/nucleic acid complex. As the Examiner notes, there

are hydrophobic interactions between individual lipids in liposomes. This hydrophobic interaction, together with partitioning the hydrophilic parts of the lipids into contact with aqueous environment and the hydrophobic parts of the lipids away from the aqueous environment causes the liposome to form. A PHOSITA would know that in the field of transfection, the cationic (hydrophilic) parts of the liposome lipids interact with the negatively charged nucleic acid. Thus, a PHOSITA would be led away from Applicants' invention because interaction of liposome lipids with a hydrophobic component of an RNA might perturb or interfere with the normal hydrophobic interaction between lipids and electrostatic interaction between lipid and the nucleic acid.

'732 contains an astonishing range of modifications and delivery vehicles which may potentially be used to delivery their siRNA. '732 also teaches a wide range of desired properties for these modifications and delivery vehicles. The authors provide no clear guidance as to which modifications, delivery vehicles, or combinations possess the desired properties. Further, there is no data of any kind to support any particular modification for combination with a delivery vehicle. The PHOSITA would understand that it is not in the realm of possibility that all modifications could be used in relation to all of the delivery vehicles or that all of the modifications would possess all the desired properties.

Applicants' invention, labile linkage of a hydrophobic group to the siRNA and then use of the hydrophobic group to in interaction with an amphipathic transfection reagent is neither understood or taught by '732 so as to be intelligently reproduced. "A prior use, in order to negative novelty, must be something more than an accidental or casual one. It must, indeed, be so far understood and practiced, or persisted in, as to contribute to the sum of human knowledge and be accessible to the public, becoming an established fact in the art." (Anthracite Separator Co. v. Pollock). For one skilled in the art to combine separately disclosed parts taught by '732 would be "purely a matter of chance and not the inevitable result of its process." (International Nickel Co. v. Ford Motor Co.) and therefore not an anticipation. There is no specific teaching in the '732 that would lead to Applicants' invention. Therefore, it is the Applicants' opinion that '732 patent is not enabling and fails to describe the Applicants' instant invention sufficiently to enable a person of ordinary skill in the art to carry out Applicants' invention.

b) Rejection under 35 U.S.C. 103(a) over Fosnaugh et al. (U.S. 2003/0143732) taken with Manoharan, M. (Biochimica et Biophysica Acta 1489, 1999: 117-130) and further evidenced by Thierry et al (US 6,110,490)

It is the Applicants' opinion that the amendments and arguments made above in response to the §102(e) rejection is sufficient to overcome the §103 rejection.

viii. Claims Appendix:

1. (previously presented) A composition for delivering an RNA to a cell comprising: a reversibly modified RNA consisting of at least one hydrophobic group having one to twenty carbon atoms covalently linked to said RNA via a labile bond cleavable under mammalian physiological conditions and an amphipathic transfection reagent wherein the reversibly modified RNA and the amphipathic transfection reagent associate to form a complex and wherein the modified RNA interacts with the amphipathic transfection reagent via hydrophobic interaction between the hydrophobic group linked to the RNA and a hydrophobic part of the amphipathic transfection reagent.

2-3. (canceled)

4. (previously presented) The composition of claim 1 wherein the hydrophobic group is linked to a ribose 2′ hydroxyl of the RNA.

5. (previously presented) The composition of claim 1 wherein the labile bond is selected from the group consisting of: a silyl ether and a maleamate.

6. (previously presented) The composition of claim 4 wherein the RNA is modified at: a single ribose 2′ hydroxyl of the RNA, more than one but not all of the ribose 2′ hydroxyls of the RNA, or all of the ribose 2′ hydroxyls of the RNA.

7-9. (canceled)

10. (previously presented) The composition of claim 1 wherein the RNA is selected from the group consisting of siRNA and microRNA.

11-12. (canceled)

13. (previously presented) The composition of claim 1 wherein the modified RNA is more resistant to nucleases than the RNA if it were not linked to the hydrophobic group.

14. (previously presented) The composition of claim 1 wherein a plurality of hydrophobic groups are attached to said RNA via labile bonds.

IX. Evidence appendix:

None

x. Related proceedings appendix:

None

Pages 1-17 are respectfully submitted,

/Kirk Ekena/

Kirk Ekena, Reg. No. 56,672
Arrowhead Madison Inc.
465 Science Drive, Suite C
Madison, WI 53711
608-316-3896

I hereby certify that this correspondence is being transmitted to the USPTO on this date: 23 Jan 2012 .

/Kirk Ekena/
Kirk Ekena